

**RESERVE COPY**  
**PATENT SPECIFICATION**



Application Date: Dec. 12, 1936. No. 34196/36.

**486,898**

Complete Specification Left: Dec. 13, 1937.

Complete Specification Accepted: June 13, 1938.

**PROVISIONAL SPECIFICATION**

**Manufacture of Substances resembling Vitamines from Plants**

I, WILFRED WILLIAM GROVES, a British Subject, of 30, Southampton Buildings, London, W.C.2, do hereby declare the nature of this invention (as communicated to me from abroad by I. G. Farbenindustrie Aktiengesellschaft, a Joint Stock Company organised according to the laws of Germany, of Frankfurt a/Main, Germany) to be as follows:—

10 The juice of certain plants, for instance, of lemon and capsicum, contains substances having properties resembling those of vitamins; the chemical structure of those substances is not yet completely understood but they are believed to belong to the flavone group and for convenience are thus referred to herein. The substances resembling dyestuffs belonging to the flavone group, for instance quercetin, are extracted with alcohol from the plant, for instance, from quercus tinctoria, and the impurities are precipitated from this solution having an acid reaction, with the aid of lead acetate, the lead combined with the quercetin being then separated from the filtered solution by the addition of hydrogen sulphide and the dissolved quercetin crystallised. The vitamin-like substances in question belonging to the flavone group are contained, for instance, in the lemon and in the capsicum in so small a quantity that they could not be separated in the necessary purity with the aid of the hitherto known processes from the substances accompanying them.

By the process of this invention the vitamin belonging to the flavone group is prepared in a pure form as follows:

40 In the solution, prepared by any extraction process with alcohol or other anhydrous solvent, the flavone to be prepared is precipitated with the hydroxide of an alkali metal or an alkaline earth metal, preferably with barium hydroxide, and the precipitate is suspended in the anhydrous solvent of the final product to be prepared and the suspension is then decomposed by introducing carbon dioxide. The flavone, the acid dissociation of which is smaller than that of carbonic acid, is thus set free and remains in the dissolved state while the

acids which are stronger than the carboxy-acid and the carbonic acid remain in the precipitate. The flavone dissolved is concentrated.

In carrying out this process there must be used a substantially anhydrous solvent for the precipitation with alkali metal hydroxide or hydroxide of alkaline earth metal because the precipitate is obtained only therein and is soluble in water. As an anhydrous solvent there may be used acetone or alcohol which need not be completely concentrated and may contain water in such a quantity as does not prevent the formation of a precipitate. The decomposition by carbonic acid must be carried out in the acid anhydrous solvent, that is, in a liquid with such a small content of water that only the flavone set free is dissolved.

The raw extract prepared from plants containing flavone is suitably at first freed in known manner from the impurities by use of heavy metal salts, for instance lead acetate. For this purpose the dissolved heavy metal salt, for instance lead acetate is added to the plant extract in an acid medium in order to cause the separation of the impurities; the heavy metal salt is then also added to the solution which has been rendered alkaline whereby the flavonate of the heavy metal is separated. This flavonate is further treated in an aqueous or anhydrous suspension.

If the precipitate in an aqueous solution is decomposed with an acid stronger than is the flavone, for instance hydrogen sulphide, the aqueous flavone solution which remains after the heavy metal salt has been separated is concentrated to such an extent that the impurities are precipitated by the addition of an anhydrous solvent and that the solution is sufficiently anhydrous for the separation of the flavone by the action of barium hydroxide. But when the precipitate of flavonate obtained by means of the lead acetate is worked up in an anhydrous suspension the flavone may directly be separated from the remaining anhydrous solution by means of barium hydroxide after the lead has been removed with an acid stronger than is the flavone.

*[Priority]*

Price 2s. 8d.

Price 2s.

Dated this 12th day of December, 1936.

ABEL & IMRAY,  
Agents for the Applicant.

# COMPLETE SPECIFICATION

## Manufacture of Substances resembling Vitamines from Plants

I, WILFRED WILLIAM GROVES, a British Subject, of 30, Southampton Buildings, London, W.C.2, do hereby declare the nature of this invention (as communicated to me from abroad by I. G. Farbenindustrie Aktiengesellschaft, a Joint Stock Company organised according to the laws of Germany, of Frankfurt a/Main, Germany) and in what manner the same is to be performed, to be particularly described and ascertained in and by the following statement:—

Certain plants, especially of the citrus species, contain glucosidic substances belonging to the flavanone group and having properties resembling those of vitamines. The chemical structure of these substances (which were formerly believed to belong to the flavone group) is not yet completely understood. The main product appears to be a hitherto unknown glucoside of eriodictyol, obtained partly as such as a substance readily soluble in water and partly as a double compound with hesperidin as a crystalline compound sparingly soluble in water.

Owing to the solubility properties of the eriodictyol glucoside the hitherto known processes for obtaining substances of this chemical group from plants, as described, for instance, by Klein, "Handbuch der Pflanzenanalyse" Vol. 3, part 2 (1932) pages 881—889, are not successful for the isolation of this compound and permit the production only of very impure final products which are not useful for pharmaceutical purposes.

By this invention the substances mentioned are prepared in a considerably purer form from their solutions obtained from the plants by any well known extraction process conducted with alcohol or other organic solvent miscible with water by precipitating the flavanone derivative from the solution by means of a hydroxide of an alkali metal or an alkaline earth metal and decomposing the precipitate so obtained by means of an acid. This decomposition may be performed in many ways. For instance, the precipitate may be suspended in an organic solvent miscible with water and decomposed by an acid, for instance by means of carbon dioxide, the alkali salt or the alkaline earth salt of the acid added separating in solid form and the flavanone derivative passing into solution. It may also be advantageous to decompose the

precipitate in an aqueous solution. The precipitation by means of an alkali hydroxide or alkaline earth hydroxide may, however, only be conducted in an organic solvent miscible with water, as the precipitate is only formed in these solvents and is soluble in water. Such organic solvents miscible with water are, for instance, acetone or alcohols. The solvent need not be completely anhydrous; it is only essential that there should not be present sufficient water to prevent the formation of a precipitate.

The crude extract prepared from plants containing the flavanone compounds is suitably first freed from the impurities in known manner by means of a solution of a heavy metal salt, for instance of lead acetate. For this purpose the dissolved heavy metal salt, for instance lead acetate, is added to the plant extract at first in an acid medium, the precipitate formed being discarded; the solution is then rendered alkaline, for instance, with ammonia, whereby the flavanone of the heavy metal is precipitated. This flavanone is further treated in an aqueous suspension or, for instance, in an alcoholic suspension. If the suspension in water is decomposed with an acid stronger than the flavanone, the aqueous flavanone solution which remains after the heavy metal salt has been separated is suitably concentrated to such an extent that the impurities are precipitated by the addition of the organic solvent and that the resulting solution is sufficiently anhydrous for the precipitation of the flavanone by the action of the alkali hydroxide or alkaline earth hydroxide. When, however, the precipitate of flavanone obtained by means of the lead acetate is worked up in suspension in an organic solvent the flavanone derivative remaining in the solution after the lead has been removed with an acid stronger than is the flavanone may be precipitated directly.

The following Examples illustrate the invention:—

1). 150 kilos of fresh lemon peels are passed through a meat-chopping machine and the comminuted material is stirred for one night together with 150 litres of alcohol of about 90 per cent strength. The following day the mass is expressed by means of a fruit press, the residue is again extracted with 75 litres of alcohol

and the combined expressed liquids are mixed with 2 per cent of barium acetate in order to remove the citric acid. The precipitated product is centrifuged and the precipitate is discarded. 1 per cent of lead acetate is then added to the clear solution, which has an acid reaction, the resulting precipitate is removed and the solution is rendered distinctly alkaline by means of ammonia. The product thus precipitated is separated by centrifuging and finely suspended in 10 litres of acetic acid of 5 per cent strength and the suspension is neutralised with ammonia, while vigorously stirring, and then strongly centrifuged. The precipitated product obtained is suspended in 10 litres of absolute alcohol and the suspension is mixed, while stirring and cooling it, with concentrated sulphuric acid until the reaction is acid to Congo paper. The precipitated product is separated and washed with alcohol and the alcoholic washings are added to the solution from which the precipitate was separated; the precipitate is discarded.

The solution is then neutralised, while vigorously stirring, by carefully adding alcoholic caustic soda solution until the reaction is neutral to Methyl Red. After the solution has been allowed to stand for some time it is decanted from the resinous precipitate. This precipitate is dissolved in 150 cc. of water, the solution is brought to a neutral reaction to Methyl Red and mixed with 1 litre of alcohol; the precipitate thus formed is removed and the liquid is combined with that decanted from the above resinous precipitate. The solution is then cooled during one night to  $-15^{\circ}\text{C}$ ., then decanted from the precipitate produced into a vessel cooled to  $-15^{\circ}\text{C}$ . and mixed, while vigorously stirring, with alcoholic caustic soda solution which is introduced in the form of a fine jet until phenol-phthalein is coloured strongly red by the solution. The precipitate is immediately centrifuged in an atmosphere of nitrogen and then taken up in N-hydrochloric acid. The solution so obtained is mixed with bicarbonate until the reaction is negative to Congo paper. When the solution is allowed to stand, a part of the flavanone derivative namely, the double compound of eriodictyol glucoside and hesperidin, separates. The remaining solution of eriodictyol glucoside is colorimetrically adjusted to a desired content, filtered under sterile conditions and filled into ampoules.

A further quantity of flavanone derivative may be obtained from the neutral precipitate obtained on cooling to  $-15^{\circ}$

C. as above described by dissolution in water, precipitation with alcohol and further treatment of the solution as described above.

2). 100 kilos of bitter orange peels freed from the outer yellow, oil-containing layer are passed through a meat-chopping machine and the comminuted mass is extracted, while heating, first with 200 litres and then with 100 litres of absolute alcohol. The combined extracts are rendered neutral to Methyl Red and then cooled. By further treatment according to the later steps described in Example 1, that is cooling to a low temperature and precipitation with alcoholic caustic soda solution, a sufficiently pure product is obtained without a precipitation with lead acetate.

Having now particularly described and ascertained the nature of my said invention and in what manner the same is to be performed, I declare that what I claim is:—

1. A manufacture of substances of the flavanone group having properties resembling those of vitamins from their solutions obtained by extraction of plant parts containing the desired substances by means of an organic solvent miscible with water, with or without purification of the crude extract so obtained, which comprises adding an alkali hydroxide or alkaline earth hydroxide to the solution, separating the precipitate and decomposing it by means of an acid.

2. A manufacture as claimed in claim 1, wherein the crude extract is first purified by adding to it at an acid reaction a heavy metal salt, for instance lead acetate, separating the precipitate from the solution, then rendering the separated solution alkaline, for instance, by addition of ammonia and removing the metal from the precipitate thus formed, the product freed from metal being then treated in solution in an organic solvent miscible with water in the manner set forth in claim 1.

3. A manufacture of substances of the flavanone group having properties resembling those of vitamins, substantially as described in either of the Examples herein.

4. Substances of the flavanone group having properties resembling those of vitamins which are obtainable by the manufacture claimed in claim 1 or claim 2, whenever prepared or produced by the process of manufacture particularly described and ascertained herein or by any process which is an obvious chemical equivalent thereof.

Dated this 13th day of December, 1937.

ABEL & IMRAY,  
Agents for the Applicant,  
30, Southampton Buildings,  
London, W.C.2.

Leamington Spa: Printed for His Majesty's Stationery Office, by the Courier Press.—1938.